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INSIDE JEB

Mayflies can't handle long, hot summers

Neocloeon triangulifer larva. Photo credit: David Funk, Stroud Water Research Center.

They don't look like your archetypal canary, but when mayfly larvae vanish from the waterways, you know the ecosystem is in trouble. 'Mayflies are often important players in freshwater ecosystems and are widely used as indicators of ecological status', says David Buchwalter, from North Carolina State University, USA, adding that the aquatic larvae, which are vulnerable to pollution and rising temperatures, can vanish without warning when conditions become harmful. However, Buchwalter was concerned about our limited understanding of the impact that high temperatures might have on these essential members of the freshwater ecosystem. 'Studies of aquatic insect thermal limits have historically been done by heating larvae until they drop', says Buchwalter. But few insects experience the steep and high temperature increases that are investigated in the lab in their natural surroundings. Concerned that scientists weren't building a realistic picture of mayfly thermal tolerance, Buchwalter decided to investigate the physiological impact of temperatures that mimic and exceed those that the larvae might genuinely experience on a hot summer's day.

Although many mayfly species are difficult to rear in the lab – the life cycle can be long, complex and often requires flowing water – David Funk, John Jackson and Bernard Sweeney from the Stroud Water Research Center, USA, had successfully isolated and reared a few species in the lab, including *Neocloeon triangulifer*, which reproduce asexually and have a much simpler life history. 'We had to determine the chronic thermal limits of this species', says Buchwalter, so Funk reared over 3000 larvae from eggs to adulthood across temperatures ranging from 14 to 30°C over several months to find out how they coped.

Although the growing larvae survived well at temperatures up to 26°C, something went drastically wrong at 28°C, when the death rate rocketed to 80%. However, when the team warmed 23-day-old larvae rapidly, they were able to cope with much higher temperatures (40°C) before succumbing to the effects. 'Insects clearly can deal with relatively warm water on a short-term basis, but cannot sustain prolonged exposures', says Buchwalter. So what was causing the insect's vulnerability?

One possibility was that the larvae simply could not supply enough oxygen for their tissues to sustain their suped-up metabolism at extremely high temperatures. If this was the case, Buchwalter reasoned that as the temperature increased, the larvae would activate genes at high temperatures that should help them to deal with a reduced oxygen supply (hypoxia), in addition to experiencing a significant reduction in the larvae's spare metabolic capacity for activities beyond those required for basic survival.

However, when Buchwalter measured the larvae's resting and maximum metabolic rates at 22, 26 and 30°C, he was surprised that their ability to provide sufficient oxygen at the highest temperatures was not compromised. And when Kyoung Sun Kim and Hsuan Chou painstakingly searched for evidence that the larvae were activating genes that would help them to deal with hypoxia as the temperatures rose, they found that the genes were only activated at temperatures that were in excess of those that the larvae naturally experience on a hot day. Whatever is killing overheated larvae on a hot summer's day, it is probably not their ability to supply enough oxygen to tissues as their metabolism rockets.

'Insect respiratory systems are very efficient and may not be limited by higher temperatures under ecologically relevant conditions', says Buchwalter, adding, 'I think we need to get away from acute thermal challenge studies and focus on environmentally relevant thermal regimes to better understand how temperature imposes limits on species'.

10.1242/jeb.165654

Kim, K. S., Chou, H., Funk, D. H., Jackson, J. K., Sweeney, B. W. and Buchwalter, D. B. (2017). Physiological responses to short-term thermal stress in mayfly (*Neocloeon triangulifer*) larvae in relation to upper thermal limits. *J. Exp. Biol.* **220**, 2598-2605.

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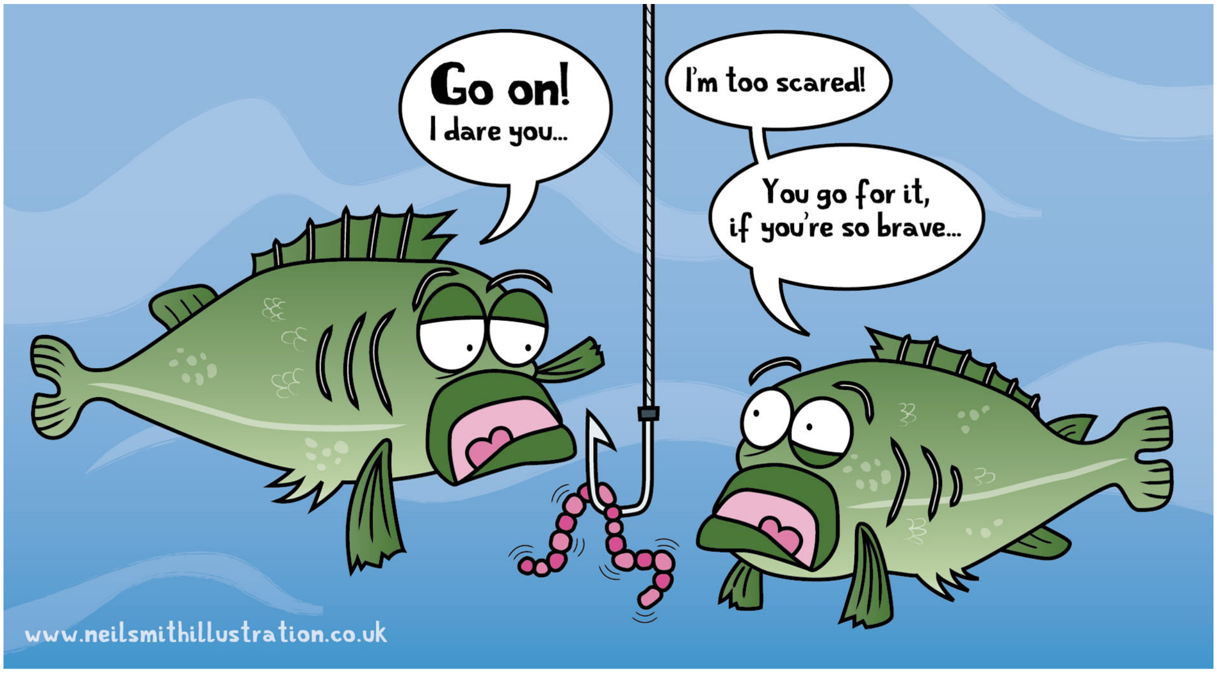
Stressed out fish are less likely to get caught



Despite the apparently idyllic setting, when a fisherman settles down beside a peaceful lake he is embarking on a battle of wills, where the ultimate reward is to catch a bite. 'I find the question of what makes a fish hit a lure to be really interesting', says Michael Louison, from the Illinois Natural History Survey, USA. 'I've done a lot of fishing and wondered why in some cases I can pitch a lure near a fish and get an immediate strike, while in other cases the fish ignores it or swims off'. Intrigued by the factors that might drive one fish to lunge while another hangs back, Louison says, 'We know in all sorts of animals that physiology drives many decisions and we wanted to know if certain behavioural or physiological traits were key in leading a fish to be more likely to attack a lure'.

Fortunately, Louison and his colleagues, Jeffery Stein and Cory Suski, had access to two populations of largemouth bass – one descended from fish that readily snapped at bait and a second descended from a population that was more reluctant to bite – which had been nurtured by the Illinois Natural History Survey since the late 1970s. However, before Louison could settle down to his favourite pastime and 'net' some fish, he and Shivani Adhikari had to assess the animals' boldness and responses to stress.

Isolating individuals from both populations in a tank divided into four zones, Louison and Adhikari monitored how eager each fish was to explore its surroundings. They then measured the amount of stress the animal experienced a day later, by holding the fish out of water for 3 min and then recording the levels of the stress hormone – cortisol – in the fish's blood. In addition, the duo tagged the fish so that they could identify each individual and measure its oxygen consumption while it was recovering from fast swimming to calculate its metabolic rate. Once Louison and Adhikari had completed the measurements, they released the animals into a pond that had been specially prepared for the study, and then Louison and Ryan Solomon collected their rods and went angling for a couple of hours a day for a week during the summer, logging each fish that they caught before returning them to the water.

However, after analysing the catch rates of the fish descended from the population that had been difficult to catch in the 1970s and those descended from the second population, which had been keen to bite and were highly vulnerable to capture, Louison was surprised that they were the same. 'We had expected that our "high vulnerability line" would actually be more likely to be caught', he says, although he suspects that they are still more catchable, but possibly at other times of year or when using different baits. The characteristic that seemed to be linked most strongly to the fish's ability to avoid capture was their response to stress. 'Fish that showed a more pronounced rise in cortisol levels after an air exposure were less likely to be caught', says Louison. He suspects that the fish that are more sensitive to stress may be shyer and less prepared to risk snapping at tasty morsels dangling before them, helping them to avoid capture.

But what could this mean for fish populations that are constantly under the fisherman's eye? 'We can definitely expect to see selection favouring fish that are harder to catch', says Louison, which could make it trickier for anglers to land fish in the future.

10.1242/jeb.165670

Louison, M. J., Adhikari, S., Stein, J. A. and Suski, C. D. (2017). Hormonal responsiveness to stress is negatively associated with vulnerability to angling capture in fish. *J. Exp. Biol.* **220**, 2529-2535.

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